Extent of digestion and rumen condition as factors affecting passage of liquid and digesta particles in sheep

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SUMMARY

Two experiments were carried out at Mt. Cotton, The University of Queensland, from November 1992 to July 1993, to study the effect of extent of digestion or feed type (grass or legume) on particle kinetics in the rumen. Small (0·5–1·18 mm) Yb-labelled grass or legume particles, either digested or undigested, were injected into the rumen of sheep fed on different diets, and their retention time in the reticulo-ruminal compartment measured. In Expt 1, four intact wethers were fed on either pangola grass hay, chaffed lucerne hay, pelleted lucerne hay or commercial pelleted concentrate. Digested particles from the faeces of animals fed on pangola or lucerne and undigested material from the same diets were wet-sieved and the fraction 0·5–1·18 mm collected, labelled with Yb-acetate and injected into the animals together with a solution of Cr-EDTA. Faecal samples were taken and analysed for marker concentrations. In Expt 2, four similar animals, fitted with duodenal and ruminal cannulae, were fed on different proportions of pangola grass hay and lucerne hay, and Cr-EDTA and the above mentioned labelled particles were injected through the rumen cannula. Samples were taken from the duodenum and analysed for marker concentrations.

The results indicated that diet characteristics rather than extent of digestion or particle type had the greatest influence on rates of passage of both liquid and particulate phases. Different proportions of pangola and lucerne did not result in marked differences in either the volumes of rumen contents or the rates of passage of the solid phase marker but altered the rates of passage of Cr-EDTA. Increasing the proportion of legume increased intake and decreased retention time markedly, with no additive effects on digestibility.

Particles of the same small size escaped with the same fractional passage rate within each diet, irrespective of type (grass or legume) or status (undigested or digested), indicating identical kinetics within each rumen type.

It was concluded that rumen conditions as influenced by diet type have most influence on water and particle kinetics and that extent of digestion of the small particles used in our experiments was not important. Particles of legume or grass of the same size behaved similarly within a diet type.

INTRODUCTION

Voluntary feed intake of low quality forages by ruminants is thought to be mainly limited by the rate of passage of digesta from the reticulo-rumen into the lower gut (Weston 1985). This parameter is highly dependent on particle size reduction (Ulyatt *et al.* 1976), and as virtually all reduction occurs prior to the omasum (Poppi *et al.* 1980, 1985; Udèn & Van Soest 1982) comminution rate has been emphasised as the dominant influence in regulating the clearance of fibrous residues from the reticulo-rumen (Ulyatt 1983). However, although the critical size for escape has been established to be *c.* 1·2 mm (Poppi *et al.* 1980), > 50% of total dry matter (DM) rumen particles are smaller (Welch 1982; Moseley & Jones 1984; Poppi *et al.* 1985), and the opening diameter of the reticulo-omasal orifice has been shown to be large enough to allow the passage of particles > 1·2 mm (McBride *et al.* 1984). Density of particles is important (DesBordes & Welch 1984; Ehle 1984) and has recently been suggested, together with extent of digestion, as one of the major factors influencing the

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rate of escape from the rumen rather than particle size *per se* (Sutherland 1988).

Legumes are introduced into pastures to increase protein supply to the intestines and their ability to do so depends, among other factors, on the escape properties of legume particles within a rumen dominated by grass particles. The extent to which rumen conditions influence the escape of these legume particles is not known.

The aim of this work was to investigate whether extent of digestion influenced escape and whether the fractional passage rate of defined labelled particles depended on rumen conditions promoted by the diet.

MATERIALS AND METHODS

Animals and diets

In Expt 1, four Merino × Dorset wethers (12 months old, mean liveweight 34·2 ± 2·14 kg at the beginning of the experiment) were fed at hourly intervals on chaffed lucerne hay (LC), pelleted lucerne hay (LP), chaffed pangola grass hay (PH) or commercial pelleted concentrate (C) offered at 90% of previously established *ad libitum* feed intake. Each animal received the four experimental diets in four different periods, following a 4 × 4 Latin square design. In Expt 2, another four animals (13 months old, 36·3 ± 1·30 kg mean liveweight), fistulated in the rumen and duodenum (T-shape cannula), were fed *ad libitum* at hourly intervals on 100% pangola hay (100P), 67% pangola hay + 33% lucerne hay (67P33L), 33% pangola hay + 67% lucerne hay (33P67L) or 100% lucerne hay (100L), according to a 4 × 4 Latin square design. All animals had free access to water.

Experiment 1

For each period of the Latin square, after 10 days adaptation to the diets the feed offered was fixed at 90% of *ad libitum* and a digestibility trial was performed for 7 days. At the end of this, four types of Yb-labelled particles of a size between 0·5 and 1·18 mm were given to the animals through an oesophageal tube, directly into the reticulo-rumen. The four types of Yb-labelled particles were digested lucerne, digested pangola, undigested lucerne and undigested pangola. Following the order of a Latin square within each period, each animal received 10 g of the four particle types in four subperiods. At the time of injection of labelled particles, 100 ml of a solution containing 1 mg Cr-EDTA/ml was also given to the animals via the oesophageal tube. Faecal samples were subsequently taken at regular intervals for up to 120 h to calculate slow (k_1) and fast (k_2) fractional outflow rates of both particulate and liquid phases according to the procedures described by Grovum & Williams (1973).

Experiment 2

For each period of the Latin square there was a 14-day adaptation to the new diets by which stage maximum intake was achieved and maintained, and a 7-day digestibility trial was then performed. Cr-EDTA was continuously infused (1 mg/ml, 100 ml/day) directly into the rumen during the last 6 days of the digestibility trial, at the end of which the infusion was stopped and duodenal samples were then taken at 0, 3, 6, 12, 18, 24 and 36 h. Yb-labelled particles as described above were then injected directly into the rumen, according to the same design used in Expt 1, and duodenal samples were collected at the same times as described for Cr-EDTA (except for time 0). Rates of passage of Cr-EDTA and Yb-labelled particles through the ruminal compartment (k_p) were then calculated. Rumen content volume was measured by manual emptying at the end of each experimental period, weighing and sampling for dry matter (DM) and chemical analysis. Apparent retention times of DM and neutral detergent fibre (NDF) in the rumen were calculated (Minson 1966).

Particle types

Faeces from animals fed on pangola or lucerne were soaked in water overnight and then mashed in a mortar and resuspended in water. Undigested particles came from diets ground through a 3 mm screen. Both digested and undigested particles were wet-sieved (Poppi *et al.* 1980) and the fraction retained on a screen pore size of 0·5 mm after passing through a screen pore size of 1·18 mm was collected. In a Latin square arrangement, all four types of particles were incubated in nylon bags suspended in the rumen of four sheep similar to the experimental animals and fed on the lucerne chaff used in Expt 1. Incubation times were 3, 6, 9, 12, 15, 18, 24, 36, 48, 72 and 96 h, and measurements of extent and rate of degradation of undigested particle DM were obtained by fitting the degradation values at each time to the model proposed by Ørskov & McDonald (1979). Digested particle data did not fit the exponential model but fitted a linear function.

Marker techniques

Cr-EDTA was prepared by the methods of Downes & McDonald (1964) and Yb-labelled particles by soaking the forages in a buffer acetate (0·1 M-acetic acid to pH 6·0 with ammonium hydroxide) for 3 h and then overnight in the same solution with an exposure of 17 mg of ytterbium acetate/kg DM. The labelled material was washed several times with distilled water and allowed to dry at 80 °C for 48 h. Faecal samples collected after marker injection were oven dried at 80 °C for 48 h, whereas duodenal material was frozen until marker analysis was per-

formed. Samples from labelled particles and faeces were analysed for marker concentrations by adding 5 ml of a 5:1 mixture of nitric and perchloric acids to *c.* 0·2 g DM of sample. The same amount of acid mixture was added to 10 g of thawed duodenal material slightly desiccated to avoid excessive acid dilution. After an overnight standing period, the samples were gradually heated until they were completely digested. Marker concentrations were determined by atomic emission spectrometry (AES-ICP).

Chemical analysis

The proportions of DM in feeds, labelled particles, faeces and ruminal and duodenal digesta were determined by drying at 100 °C, and organic matter (OM) by ashing at 550 °C. Total nitrogen (N) was determined following the Kjeldahl method, ammonia content of ruminal digesta by distillation with $Na_2B_4O_7$, and NDF was measured on dried samples according to the method of Goering & Van Soest (1970).

Statistical analysis

Voluntary feed intake, digestibility and rumen degradation kinetics of 0·5–1·18 mm particles (Expts 1 and 2), as well as volume of rumen contents and Cr-EDTA outflow rate from rumen (Expt 2) were analysed by analysis of variance following the methods proposed by Steel & Torrie (1980) and according to the model:

$$y_{ijkl} = \mu + D_i + A_j + P_k + \epsilon_{l(ijk)}$$

where D_i (D.F. = 3) represents the effect of the diet, A_j (D.F. = 3) the effect due to the animals, P_k (D.F. = 3) the period effect and $\epsilon_{l(ijk)}$ (D.F. = 6) the experimental error. All effects were compared to $\epsilon_{l(ijk)}$. There was one missing value of Cr-EDTA outflow rate, which was estimated as appropriate. The analysis of variance was performed in the usual manner with one D.F. being subtracted from error.

Slow (k_1) and fast (k_2) fractional outflow rates of both Yb-labelled particles and Cr-EDTA (Expt 1) as well as fractional outflow rate determined from duodenal samples (k_P) of Yb-labelled particles (Expt 2) were analysed following the model:

$$y_{ijklmn} = \mu + D_i + A_j + P_k + S_{l(k)} + I_m + DI_{im} + AI_{jm} + PI_{km} + \epsilon_{n(ijklm)}$$

where D_i (D.F. = 3) represents the effect of the diet, A_j (D.F. = 3) the effect due to the animals, P_k (D.F. = 3) the period effect, $S_{l(k)}$ (D.F. = 12) the effect due to the subperiods of infusion of different Yb-labelled particles within each dietary period, I_m (D.F. = 3) the effect due to the kind of labelled particles infused, DI_{im} (D.F. = 9) the interaction between diet and kind of infused labelled particles, AI_{jm} (D.F. = 9) the interaction between animals and infused particles, PI_{km} (D.F. = 9) the interaction between dietary periods and infused particles and $\epsilon_{n(ijklm)}$ (D.F. = 12) the experimental error. The interaction between animals and type of particles infused (AI_{jm}) was used as the error term to test for particle type effects (I_m). All other effects were estimated and tested from $\epsilon_{n(ijklm)}$. In Expt 2 there were ten missing values of Yb-labelled particles outflow rate, which were estimated by considering the Latin squares arranged for particles infusion within each dietary period as independent. Analysis of variance was performed as usual, subtracting ten D.F. from error.

Where the effect studied was significant, mean values were compared by the least significant difference test, taking into account missing data when needed.

The sums of squares of significant factors from Expt 2 were partitioned in three orthogonal contrasts to test linear, quadratic and cubic responses to legume inclusion in the diet.

RESULTS

Chemical composition, intake and digestibility of the diets

The chemical composition of the diets is given in Table 1, and intake and digestibility in Tables 2 (Expt 1) and 3 (Expt 2). Pangola had lower crude protein and higher NDF than lucerne, as expected.

In Expt 1, DM and OM digestibility values were significantly higher ($P < 0·01$) for diet C, whereas CP digestibility was low ($P < 0·001$) for diet PH (Table 2). Diet LP had the lowest NDF digestibility ($P < 0·05$) whereas PH had the highest (Table 2). Values were in the expected range. Differences between diets for digestible organic matter intake (DOMI, g/kg $W^{0·75}$ per day) were less than those for DM intake (DMI).

In Expt 2 there was a significant animal effect ($P < 0·05$) for both DMI and DOMI. The DM, OM and CP digestibility coefficients were positively and lin-

Table 1. *Chemical composition of the diets as dry matter* (*DM*), *organic matter* (*OM*), *crude protein* (*CP*) *and neutral detergent fibre* (*NDF*). *In Expt 1, concentrate* (*C*), *pangola hay* (*PH*), *lucerne chaff* (*LC*) *and lucerne pellets* (*LP*) *were fed, whereas in Expt 2, combinations of pangola hay and lucerne chaff were fed*

	Expt 1				Expt 2	
	C	PH	LC	LP	PH	LC
DM (g/kg)	884	896	884	885	882	851
OM (g/kg DM)	885	934	898	906	939	897
CP (g/kg DM)	200	60	178	179	53	227
NDF (g/kg DM)	345	721	495	500	716	403

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Table 2. *Dry matter intake* (*DMI*) *and digestible organic matter intake* (*DOMI*) (*g/kg* $W^{0.75}$ *per day*), *and digestibility coefficients* (%) *of dry matter* (*DMD*), *organic matter* (*OMD*), *crude protein* (*CPD*) *and neutral detergent fibre* (*NDF*) *of the diets used in Expt 1* (*C*, *concentrate; PH*, *pangola hay; LC*, *lucerne chaff and LP*, *lucerne pellets*). *Mean values for four sheep*

	Diet				
	C	PH	LC	LP	S.E. (6 D.F.)
DMI	75·3	36·9	68·4	118·2	6·40
DMD	68·2	51·0	56·5	52·4	1·64
OMD	71·6	51·9	57·9	53·5	1·59
CPD	80·8	30·7	71·9	67·0	3·87
NDFD	46·5	52·0	42·9	37·2	2·28
DOMI	47·9	17·8	35·5	54·1	4·00

Table 3. *Dry matter intake* (*DMI*) *and digestible organic matter intake* (*DOMI*) (*g/kg* $W^{0.75}$ *per day*), *and digestibility coefficients* (%) *of dry matter* (*DMD*), *organic matter* (*OMD*), *crude protein* (*CPD*) *and neutral detergent fibre* (*NDF*) *of the diets used in Expt 2* (*100P*, *100% pangola; 67P33L*, *67% pangola and 33% lucerne; 33P67L*, *33% pangola and 67% lucerne*, *and 100L*, *100% lucerne*). *Mean values for four sheep*

	Diet				
	100P	67P33L	33P67L	100L	S.E. (6 D.F.)
DMI	29·8	59·8	70·2	83·1	4·62
DMD	55·9	60·5	65·1	66·3	2·59
OMD	57·2	61·7	66·4	67·9	2·64
CPD	17·3	69·1	78·2	82·2	3·15
NDFD	57·3	53·1	50·3	47·3	2·88
DOMI	16·0	34·1	42·5	51·0	2·62

early related to the proportion of legume in the diet. There were no significant differences between diets for the NDF digestibility coefficient. The experimental period had a tendency to be significant ($P < 0{\cdot}1$). Differences between diets for DOMI were less than for DMI. Only the linear components of the regressions of the various parameters studied on legume proportion in the diet were significant, except for CP digestibility, to which both quadratic and cubic components were significant ($P < 0{\cdot}01$).

Degradation characteristics of the 0·5–1·18 mm particles incubated in nylon bags are shown in Table 4. There were significant differences between undigested and digested particles for both extent ($P < 0{\cdot}001$) and rate ($P < 0{\cdot}01$) of degradation. No differences ($P > 0{\cdot}05$) were found in the extent of digestion that were due to the origin of particles (grass or legume) for either undigested or digested particles. However, the fractional rate of degradation was faster for legume particles in both instances ($P < 0{\cdot}001$ for undigested particles and $P < 0{\cdot}05$ for digested particles).

Table 4. *Potential extent* ($a+b$; %) *and fractional rate* (c; h^{-1}) *of degradation in nylon bags of the unlabelled 0·5–1·18 mm particles injected in Expts 1 and 2* (*DL*, *digested lucerne particles; DP*, *digested pangola particles; UL*, *undigested lucerne particles; UP*, *undigested pangola particles*). *Mean values for four sheep*

	Type of particle				
	DL	UL	DP	UP	S.E. (6 D.F.)
$a+b$	3·1	49·0	6·0	57·7	2·58
c	0·019	0·067	0·012	0·027	0·0019

Rates of passage of Cr-EDTA and Yb-labelled particles

Experiment 1

The k_1 and k_2 values for Cr-EDTA were affected by both diet type (Table 5) and the experimental period ($P < 0{\cdot}05$). However, there were differences between animals for k_1 ($P < 0{\cdot}05$) but not for k_2.

For the Yb-labelled particles there were differences between animals ($P < 0{\cdot}05$) and periods ($P < 0{\cdot}01$) for k_1 but not for k_2 values, whereas diet and thus rumen type affected both k_1 and k_2 (Table 5). The type of particle (grass or legume) and digested status did not influence k_1 or k_2 (Table 5). The mean values for digested status were 0·0405 *v.* 0·0403 h^{-1} for digested *v.* undigested particles, and for origin of particles 0·0406 *v.* 0·0402 h^{-1} for lucerne *v.* pangola. Moreover there was no significant interaction between diet and type of particles for k_1 and k_2.

Experiment 2

In this experiment, Cr-EDTA k_p was significantly affected by diet type (Table 6). Only the linear component of the regression of k_p on lucerne proportion in the diet was significant ($P < 0{\cdot}001$). For the Yb-labelled particles differences in k_p due to either diet or type of infused particles (Table 6) were not significant ($P > 0{\cdot}05$).

Rumen contents

The weight of the rumen contents in animals from Expt 2 was not significantly affected by diet type (Table 7). There was a significant animal effect on the weights of all DM fractions, except the NDF. Apparent retention time of DM and NDF was significantly affected by diet type. Linear, but not

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Table 5. *Expt 1, slow* (k_1) *and fast* (k_2) *fractional outflow rates* (h^{-1}) *of Cr-EDTA and Yb-labelled small (0·05–1·18 mm) particles as affected by diet (C, concentrate; PH, pangola hay; LC, lucerne chaff and LP, lucerne pellets) and particle type (DL, digested lucerne particles; DP, digested pangola particles; UL, undigested lucerne particles; UP, undigested pangola particles). Cr-EDTA values relate to the periods when the different particles were injected. Mean values for four sheep*

	Diet					
	C	PH	LC	LP	Mean	S.E.[1](9 D.F.)
Cr-EDTA						
k_1						
DL	0·046	0·040	0·058	0·109	0·063	0·0030
UL	0·053	0·042	0·063	0·099	0·064	
DP	0·050	0·046	0·066	0·095	0·064	
UP	0·045	0·039	0·067	0·094	0·061	
Mean	0·049	0·042	0·064	0·099	——	
S.E.[2] (12 D.F.)			0·0031			
k_2						
DL	0·195	0·102	0·168	0·428	0·222	0·0526
UL	0·139	0·110	0·284	0·480	0·253	
DP	0·128	0·139	0·405	0·459	0·283	
UP	0·094	0·101	0·447	0·396	0·259	
Mean	0·139	0·113	0·325	0·441	——	
S.E.[2] (12 D.F.)			0·0457			
Yb-labelled						
k_1						
DL	0·030	0·028	0·044	0·055	0·039	0·0018
UL	0·036	0·031	0·042	0·058	0·042	
DP	0·025	0·035	0·054	0·053	0·042	
UP	0·024	0·031	0·049	0·051	0·039	
Mean	0·029	0·031	0·047	0·054	——	
S.E.[2] (12 D.F.)			0·0022			
k_2						
DL	0·140	0·073	0·115	0·253	0·145	0·0233
UL	0·086	0·080	0·175	0·180	0·130	
DP	0·086	0·067	0·156	0·335	0·161	
UP	0·089	0·067	0·118	0·179	0·114	
Mean	0·100	0·072	0·141	0·237	——	
S.E.[2] (12 D.F.)			0·0200			

[1] S.E. for comparisons between particle types.
[2] S.E. for comparisons between diets.

quadratic or cubic, components of the regressions of the different parameters on legume proportion in the diet were significant.

DISCUSSION

Chemical composition, intake and digestibility of diets were within the expected range of values for these diets. When pangola and lucerne DMI were compared between the two experiments, there were lower intakes for pangola (24%) and higher for lucerne (21%) in Expt 2. However, when intake was expressed on a DOM basis, differences were less for pangola (11%) but increased for lucerne (44%), although as a result of its better quality in Expt 2 lucerne NDF intake was similar to that in Expt 1 (33·9 *v.* 33·5 g/kg $W^{0·75}$ per day). As expected, digested particles had a much lower potential for digestion than undigested particles (Table 4) and although incubation of Yb-labelled particles was not performed, it is highly unlikely that the labelling process would affect the differences in potential digestibility of the two types of particles.

These two experiments were done to examine the effect of particle source (grass or legume), extent of digestion of particle (digested or undigested) and rumen conditions as influenced by diet on kinetics of Yb-labelled particles and Cr-EDTA in the rumen. This study has examined a much wider range of diet types and grass/legume combinations than is usual,

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Table 6. *Expt 2, rumen fractional outflow rates (h^{-1}) of Cr-EDTA and small (0·5–1·18 mm) Yb-labelled particles, determined by analysing duodenal digesta, as affected by the diet (100P, 100% pangola; 67P33L, 67% pangola and 33% lucerne; 33P67L, 33% pangola and 67% lucerne, and 100L, 100% lucerne) and the type of particle (DL, digested lucerne particles; DP, digested pangola particles; UL, undigested lucerne particles; UP, undigested pangola particles). Mean values were corrected for missing data*

	Diet						
	100P	66P33L	33P66L	100L	Mean	R.S.D.	D.F.
Cr-EDTA	0·037	0·053	0·060	0·079	——	0·0065	5*
Yb-labelled							
DL	0·044	0·031	0·040	0·037	0·036		
UL	0·027	0·026	0·038	0·034	0·032	0·0130†	9
DP	0·039	0·033	0·038	0·052	0·041		
UP	0·023	0·025	0·021	0·032	0·025		
Mean	0·030	0·029	0·034	0·039			
R.S.D.		0·0330‡					
D.F.		2§					

* One missing value.
† R.S.D. for comparisons between particle types.
‡ R.S.D. for comparisons between diets.
§ Ten missing values.

Table 7. *Expt 2, rumen contents of whole digesta (WD), dry matter (DM), organic matter (OM), ammonia nitrogen (NH_3-N) and neutral detergent fibre (NDF), and apparent retention time of DM (RTDM) and NDF (RTNDF) in the rumen as affected by the diet of varying percentages of pangola (P) and lucerne (L). Mean values for four sheep*

	Diet				
	100P	67P33L	33P67L	100L	S.E. (6 D.F.)
WD (g)	5799	6304	6152	4912	556·3
DM (g)	667	830	762	607	70·0
DM (g/kg W)	21	25	21	16	2·1
OM (g)	609	758	680	533	64·4
NH_3-N (mg)	781	892	1053	881	89·4
NDF (g)	475	575	505	384	48·3
RTDM (h)	41	24	18	12	3·2
RTNDF (h)	42	30	26	20	4·3

and the results clearly show diet, and therefore rumen conditions as influenced by diet, as having the major effect on outflow rate from the rumen of both Yb-labelled particles and Cr-EDTA.

Diet type has been shown to be important (Poppi *et al.* 1981*a* ; Ellis *et al.* 1984; Moseley & Jones 1984; Waghorn *et al.* 1986; Deswysen 1987) but it is not usual to use a common labelled particle type across all diets as was done here. This study is extensive because of the four particle types and Cr-EDTA used across all eight diets. Various reasons have been proposed for the influence of diet type, including particle size (Poppi *et al.* 1981*b*), extent of digestion (Sutherland 1988) and particle shape (Welch 1982). In this study these parameters were varied except for particle size, which was set at particles retained on a 0·5 mm sieve after passing a 1·18 mm screen, allowing their ready passage from the rumen (Poppi *et al.* 1980). Particle shape was varied by choosing legume or grass particles and extent of digestion by choosing faecal or feed particles.

Legume or grass particles had similar rumen outflow rate values irrespective of diet type. The shape of the legume particles was not determined but they would be expected to be a mixture of round disintegrated leaf particles and long stem particles (Brazle & Harbers 1977; Moseley & Jones 1984). It may be speculated that this might have reduced the

difference between grass and legume. Despite this, the two particle sources behaved similarly in both experiments, indicating that for a given size the origin of the particle (i.e. legume or grass) was unimportant. This had not been observed previously, as legumes always had a faster fractional outflow rate from the rumen, but this was confounded by diet type influencing rumen conditions (Hendricksen *et al.* 1981; Poppi *et al.* 1981*b*; McLeod *et al.* 1990).

Degradation rates determined by nylon bags are used in conjunction with passage rates, either assumed or derived from marker studies, to estimate protein supply to the animal. The results of these experiments have shown that diet type influences the passage value for a particular particle type in that degradation of protein from a legume particle is likely to be different if that particle is placed in a grass-dominated rumen or a legume-dominated rumen.

Extent of digestion also had no effect on particle kinetics in either Expt 1 or Expt 2. The two experiments examined outflow rate from the rumen by different sampling sites (faeces and duodenum, respectively) but this did not influence the results, probably because the ratio k_1:k_2 in Expt 1 was sufficiently large to allow an accurate estimation of rumen outflow rates from faecal marker excretion curves (Cruickshank *et al.* 1989). Sutherland (1988) has elegantly described the mechanism whereby extent of digestion might be important as it relates to gas entrapment and flotation. However, it appears that in our experiments Sutherland's concepts do not hold, as there were no differences ($P > 0{\cdot}05$) between undigested and digested particles. Although effects of specific gravity on particle outflow rate from the rumen have been noted before (Kaske & Engelhardt 1990; Poncet 1991), small particles which have been in the rumen for short periods of time have a density in excess of 1·1–1·2 g/ml (Nocek & Kohn 1987) and thus can readily escape through the reticulo-omasal orifice (Mathison *et al.* 1995).

The rates at which the forages eaten by ruminants are degraded or physically removed from the rumen largely determine not only the release in the rumen of nutrients for rumen microbes and the host but also the amount of forage that can be eaten; thus their knowledge is central to an understanding of the performance of ruminant animals (Faichney 1986).

There was not a good relationship between Yb k_1 (average values for the four types of particles within each diet) and intake:

Yb k_1 (%/h) =

$2{\cdot}15 + 0{\cdot}023$ DMI (g/kg $W^{0{\cdot}75}$ per day); $r^2 = 0{\cdot}48$

This probably arises because rumen-fill was not constant. Rumen-fill was measured only in Expt 2, where no significant differences were recorded. Differences would be expected in Expt 1 because of the contrasting diet types used.

There was a strong correlation between Cr k_1 and Yb k_1 for all diets. The relationship was

Yb k_1 (%/h) = $1{\cdot}27 + 0{\cdot}40$ Cr k_1 (%/h); $r^2 = 0{\cdot}74$

and demonstrated the importance of water turnover in influencing particle turnover with diet having most influence on Cr k_1 (Tables 5 and 6). This has been discussed by Faichney *et al.* (1980/81) and Faichney (1986). The precise mechanism is yet to be elucidated, although osmolarity and factors influencing final volume of rumen contents play an important role. Diet type had most influence on both Cr k_1 and Yb k_1 irrespective of particle type. The k_1 value for Yb particle type was not a characteristic of the particle but a characteristic of the rumen in which the particle was placed. Unlike fractional digestion rate, which is a characteristic of the particle with values independent of pool size, fractional passage rate is more a characteristic of the rumen conditions (pool size, water turnover) than particle type, although particle type is still ranked the same irrespective of rumen type. Caution must thus be exercised when comparing fractional outflow rates of particle types (e.g. supplements), and information on diet types or rumen conditions must be taken into account.

Expt 2 provided the opportunity to examine the effects of legume inclusion on intake, retention time and digestibility. Intake increased markedly as legume inclusion increased. Minson (1985) suggested that most of the increase in intake occurred with *c.* 30% inclusion of legume and this also occurred here although intake was linearly, not curvilinearly, related to the proportion of lucerne in the diet (Table 3). There was no consistent effect on rumen DM or NDF load but a significant effect on retention time (Table 7). There was, however, a strong linear correlation between DM or NDF intake (g/day) and their apparent retention times as determined by Minson (1966).

DM intake (g/day)

$= 1577 - 29{\cdot}0$ Retention time of DM (h); $r^2 = 0{\cdot}99$

NDF intake (g/day)

$= 695 - 9{\cdot}0$ Retention time of NDF (h); $r^2 = 0{\cdot}78$

This linear relationship would probably result from differences in the rate at which a higher proportion of particles (legume particles) reached smaller sizes, a higher rate and extent of digestion of legume particles and differences in water turnover, rather than the type of particle *per se* (Table 6). This is suggested because there was no difference between type of particle (grass or legume) but there was a difference between extent of digestion and the rumen types (Table 6).

The predicted values for OM digestibility of the combinations of grass and legume determined mathematically from the proportions and the individual grass and legume digestibility values (60·7 and 64·4%

for 67P33L and 33P67L, respectively) agreed with those observed experimentally (61·7 and 66·4%, Table 3). This indicates no major associative effects when a grass of low quality is given in combination with a legume of high quality.

Diet effect had a major influence on k_2 of Cr-EDTA and Yb particles in Expt 1, with no major effects due to particle type (Table 5). Data from Expt 2 were not available, as k_2 was not measured in this experiment. The fast fractional outflow rates were much higher than in the rumen, as expected, and a strong relationship between values from Cr-EDTA and Yb existed, illustrating once again the importance of water turnover in a pool on particle turnover.

$$\text{Yb } k_2\ (\%/\text{h}) = 0{\cdot}025 + 0{\cdot}443\ \text{Cr } k_2\ (\%/\text{h});\ r^2 = 0{\cdot}92$$

It was concluded that rumen conditions as influenced by diet type had most influence on fractional outflow rate of particles of a common size but varying in origin (grass or legume) and extent of digestion (digested or undigested). Neither extent of digestion nor origin of particles (grass or legume) had an influence on fractional outflow rate.

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REFERENCES

Brazle, F. K. & Harbers, L. H. (1977). Digestion of alfalfa hay observed by scanning electron microscopy. *Journal of Animal Science* **45**, 506–512.

Cruickshank, G. J., Poppi, D. P. & Sykes, A. R. (1989). Theoretical considerations in the estimation of rumen fractional outflow rate from various sampling sites in the digestive tract. *British Journal of Nutrition* **62**, 229–239.

DesBordes, C. K. & Welch, J. G. (1984). Influence of specific gravity on rumination and passage of indigestible particles. *Journal of Animal Science* **59**, 470–475.

Deswysen, A. G. (1987). Forestomach: control of digesta flow. In *Physiological and Pharmacological Aspects of the Reticulo-rumen* (Eds L. A. A. Ooms, A. D. Degryse & A. S. J. P. A. M. van Miert), pp. 133–154. Dordrecht, The Netherlands: Martinus Nijhoff Publishers.

Downes, A. M. & McDonald, I. W. (1964). The chromium-51 complex of ethylenediamine tetraacetic acid as a soluble rumen marker. *British Journal of Nutrition* **18**, 153–162.

Ehle, F. R. (1984). Influence of feed particle density on particulate passage from rumen of Holstein cow. *Journal of Dairy Science* **67**, 693–697.

Ellis, W. C., Matis, J. H., Pond, K. R., Lascano, C. E. & Telford, J. P. (1984). Dietary influences on flow rate and digestive capacity. In *Herbivore Nutrition in the Subtropics and Tropics* (Eds M. C. Gilchrist & R. I. Mackie), pp. 269–293. Craighall, South Africa: The Science Press (Pty) Ltd.

Faichney, G. J. (1986). The kinetics of particulate matter in the rumen. In *Control of Digestion and Metabolism in Ruminants* (Eds L. P. Milligan, W. L. Grovum & A. Dobson), pp. 173–195. Englewood Cliffs, NJ: Prentice-Hall.

Faichney, G. J., Beever, D. E. & Black, J. L. (1980/1981). Prediction of the fractional rate of outflow of water from the rumen of sheep. *Agricultural Systems* **6**, 261–268.

Goering, H. K. & Van Soest, P. J. (1970). *Forage Fiber Analysis*, *USDA Agriculture Research Service No*. 379. Washington, DC: ARS/USDA.

Grovum, W. L. & Williams, V. J. (1973). Rate of passage of digesta in sheep. 4. Passage of marker through the alimentary tract and the biological relevance of rate constants derived from the changes in concentration of marker in faeces. *British Journal of Nutrition* **30**, 313–329.

Hendricksen, R. E., Poppi, D. P. & Minson, D. J. (1981). The voluntary intake, digestibility and retention time by cattle and sheep of stem and leaf fractions of a tropical legume (*Lablab purpureus*). *Australian Journal of Agricultural Research* **32**, 389–398.

Kaske, M. & Engelhardt, W. v. (1990). The effect of size and density on mean retention time of particles in the gastrointestinal tract of sheep. *British Journal of Nutrition* **63**, 457–465.

Mathison, G. W., Okine, E. K., Vaage, A. S., Kaske, M. & Milligan, L. P. (1995). Current understanding of the contribution of the propulsive activities in the forestomach to the flow of digesta. In *Ruminant Physiology*: *Digestion*, *Metabolism*, *Growth and Reproduction* (Eds W. v. Engelhardt, S. Leonard-Marek, G. Breves & D. Giesecke), pp. 23–41. Stuttgart, Germany: Ferdinand Enke Verlag.

McBride, B. W., Milligan, L. P. & Turner, B. V. (1984). Endoscopic observations of digesta transfer from the reticulo-rumen to omasum of cattle. *Canadian Journal of Animal Science* **64**, *Supplement*, 84–85.

McLeod, M. N., Kennedy, P. M. & Minson, D. J. (1990). Resistance of leaf and stem fractions of tropical forage to chewing and passage in cattle. *British Journal of Nutrition* **63**, 105–119.

Minson, D. J. (1966). The apparent retention of food in the reticulo-rumen at two levels of feeding by means of an hourly feeding technique. *British Journal of Nutrition* **20**, 765–773.

Minson, D. J. (1985). Nutritional value of tropical legumes in grazing and feeding systems. In *Forage Legumes for Energy-Efficient Animal Production* (Eds R. F. Barnes, P. R. Ball, R. W. Brougham, G. C. Marten & D. J. Minson), pp. 192–196. Springfield, Va, US: National Technical Information Service.

Moseley, G. & Jones, J. R. (1984). The physical digestion of perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*) in the foregut of sheep. *British Journal of Nutrition* **52**, 381–390.

Nocek, J. E. & Kohn, R. A. (1987). Initial particle form and size on change in functional specific gravity of alfalfa and timothy hay. *Journal of Dairy Science* **70**, 1850–1863.

Ørskov, E. R. & McDonald, I. (1979). The estimation of protein degradability in the rumen from incubation

measurements weighted according to rate of passage. *Journal of Agricultural Science*, *Cambridge* **92**, 499–503.

Poncet, C. (1991). The outflow of particles from the reticulo-rumen. In *Rumen Microbial Metabolism and Ruminant Digestion* (Ed J. P. Jouany), pp. 297–322. Paris: INRA.

Poppi, D. P., Norton, B. W., Minson, D. J. & Hendricksen, R. E. (1980). The validity of the critical size theory for particles leaving the rumen. *Journal of Agricultural Science*, *Cambridge* **94**, 275–280.

Poppi, D. P., Minson, D. J. & Ternouth, J. H. (1981*a*). Studies of cattle and sheep eating leaf and stem fractions of grasses. I. The voluntary intake, digestibility and retention time in the reticulo-rumen. *Australian Journal of Agricultural Research* **32**, 99–108.

Poppi, D. P., Minson, D. J. & Ternouth, J. H. (1981*b*). Studies of cattle and sheep eating leaf and stem fractions of grasses. III. The retention time in the rumen of large feed particles. *Australian Journal of Agricultural Research* **32**, 123–137.

Poppi, D. P., Hendricksen, R. E. & Minson, D. J. (1985). The relative resistance to escape of leaf and stem particles from the rumen of cattle and sheep. *Journal of Agricultural Science*, *Cambridge* **105**, 9–14.

Steel, R. G. D. & Torrie, J. H. (1980). *Principles and Procedures of Statistics*. New York: McGraw Hill Book Company.

Sutherland, T. M. (1988). Particle separation in the forestomachs of sheep. In *Comparative Aspects of Physiology of Digestion in Ruminants* (Eds A. Dobson & M. Dobson), pp. 271–290. New York: Cornell University Press.

Udèn, P. & Van Soest, P. J. (1982). The determination of digesta particle size in some herbivores. *Animal Feed Science and Technology* **7**, 35–44.

Ulyatt, M. J. (1983). Plant fibre and regulation of digestion in the ruminant. In *Fibre in Human and Animal Nutrition* (Eds G. Wallace & L. Bell), pp. 103–107. Wellington, NZ: The Royal Society of New Zealand.

Ulyatt, M. J., Baldwin, R. L. & Koong, L. J. (1976). The basis of nutritive value – a modelling approach. *Proceedings of the New Zealand Society of Animal Production* **36**, 140–149.

Waghorn, G. C., Reid, C. S. W., Ulyatt, M. J. & John, A. (1986). Feed comminution, particle composition and distribution between the four compartments of the stomach in sheep fed chaffed lucerne hay at two feeding frequencies and intake levels. *Journal of Agricultural Science*, *Cambridge* **106**, 287–296.

Welch, J. G. (1982). Rumination, particle size and passage from the rumen. *Journal of Animal Science* **54**, 885–894.

Weston, R. H. (1985). The regulation of feed intake in herbage-fed ruminants. *Proceedings of the Nutrition Society of Australia* **10**, 55–62.